

SEQ ID NO:1:

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GCCCCAGTTTGAGAAAACCTCTGGAACAACTCCTAGGGGCTACCTCACTTCCTTTGA
GATGTTTAAACAGCACTTATAAGCTCTATACACATAGTTACCTGGGATTTGGATTGAA
AGCTGCAAGACTAGCAACCCTGGGAGCCCTGGAGACAGAAGGGACTGATGGGCACA
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SEQ ID NO:2:

GCGGGCTGCCGCGCAAGGGTGGCGCGCGCGCGTTCCTTGTTCTGGTCAACAAAG
AAATGTGGAGTGTCTTGGCTGAATCCTCATACAGACAAGATCATTATGGTGCTGTTA
GGTTGAAAAAGTGATATAATAAAGGAACCAAGGAGAAAATTCAGAAGGAAAGAAA
AAATTGCCTCTGCAGGTGTGCGAGCAGGATTGCTTCTGCAACAAAAGCCTCCACCCA
GCCACATCTTGGGAAAAGAATGGCCACTTCTTGGGGCACAGTCTTTTTCATGCTGGT
GGTATCCTGTGTTTGCAGCGCTGTCTCCACAGGAACCAGCAGACTTGTTTTGAGGG
TATCTTCCTGTCTTCCATGTGCCCCATCAATGTCAGCGCCAGCACCTTGTATGGAATT
ATGTTTGATGCAGGGAGCACTGGAACCTCGAATTCATGTTTACACCTTTGTGCAGAAA
ATGCCAGGACAGCTTCCAATTCTAGAAGGGGAAGTTTTTGATTCTGTGAAGCCAGGA
CTTTCTGCTTTTGTAGATCAACCTAAGCAGGGTGCTGAGACCGTTCAAGGGCTCTTA
GAGGTGGCCAAAGACTCAATCCCCGAAGTCACTGGAAAAAGACCCCAAGTGGTCCT
AAAGGCAACAGCAGGACTACGCTTACTGCCAGAACACAAAGCCAAGGCTCTGCTCT
TTGAGGTAAAGGAGATCTTCAGGAAGTCACTTTCTGGTACCAAAGGGCAGTGTTA
GCATCATGGATGGATCCGACGAAGGCATATTAGCTTGGGTACTGTGAATTTCTGA
CAGGTCAGCTGCATGGCCACAGACAGGAGACTGTGGGGACCTTGGACCTAGGGGGA
GCCTCCACCCAAATCACGTTCTTCCCCAGTTTGAGAAAACCTCTGGAACAACTCCT
AGGGGCTACCTCACTTCCTTTGAGATGTTTAAACAGCACTTATAAGCTCTATACACAT
AGTTACCTGGGATTTGGATTGAAAGCTGCAAGACTAGCAACCCTGGGAGCCCTGGA
GACAGAAGGGACTGATGGGCACACTTTCCGGAGTGCCTGTTTACCGAGATGGTTGG
AAGCAGAGTGGATCTTTGGGGGTGTGAAATACCAGTATGGTGGCAACCAAGAAGGG
GAGGTGGGCTTTGAGCCCTGCTATGCCGAAGTGCTGAGGGTGGTACGAGGAAAAC
TCACCAGCCAGAGGAGGTCCAGAGAGGTTCTTCTATGCTTTCTCTTACTATTATGA
CCGAGCTGTTGACACAGACATGATTGATTATGAAAAGGGGGGTATTTTAAAAGTTGA
AGATTTTGAAAGAAAAGCCAGGGAAGTGTGTGATAACTTGGAAAACCTTCACCTCAG
GCAGTCCTTTCTGTGCATGGATCTCAGCTACATCACAGCCCTGTTAAAGGATGGCT
TTGGCTTTGCAGACAGCACAGTCTTACAGCTCACAAAGAAAGTGAACAACATAGAG
ACGGGCTGGGCCTTGGGGGCCACCTTTCACCTGTTGCAGTCTCTGGGCATCTCCCAT
TGAGGCCACGTACTTCCTTGGAGACCTGCATTTGCCAACACCTTTTAAAGGGGAGGA
GAGAGCACTTAGTTTCTGAACTAGTCTGGGGACATCCTGGACTTGAGCCTAGAGATT
WRGTTAATTAASCGGCCGAGCTTATCCTTWATRAGGTAATTTACTTGCMTGGCCGCG
TTTACACGTCGTGATGGNAACCTGCGTCCCAACTAACGCTTGASAMATCCCCTTCG
CAGCTGCGATACCAAAGCCGACGACGCCTTCCACAGTGCCA

Figure 1

SEQ ID NO:3:

MATSWGTVFFMLVVSCVCSAVSHRNQQTWFEGIFLSSMCPINVSASTLYGIMFDAGSTG
TRIHVYTFVQKMPGQLPILEGEVFDSEVKPGLSAFVDQPKQGAETVQGLLEVAKDSIPRSH
WKKTPVVLKATAGLRLLEPHKAKALLFEVKEIFRKSPFLVPKGSVSIMDGSDEGILAWV
TVNFLTGQLHGHRQETVGTDLGGASTQITFLPQFEKTLEQTPRGYLTSFEMFNSTYKLY
THSYLGFGLKAARLATLGALETEGTDGHTFRSACLPRWLEAEWIFGGVKYQYGGNQEG
EVGFEP CYAEVLRVVRGKLHQPEEVQRGSFYAFSYYYDRAVDTDMIDYEKGGILKVED
FERKAREVCDNLENFTSGSPFLCMDLSYITALLKDGFGFADSTVLQLTKKVNNIETGWA
LGATFHLLQSLGISH

Figure 2

CD39Human.seq	M E D T R E S N V K T F C S K N I C A I L G F S S T A V I A L L A - - V C I T	38
246 prot	M A T S W G T V F - - - - - F M L V V S C V C S A V S H R N Q Q T W F E Q T F	34
CD39Human.seq	Q N K - - - - A L P E N Y K Y G I V L D A G S S H T S L Y I Y K W P A E K E N D	74
246 prot	L S S M C F I N V S A S T L Y G I M F D A G S T G T R I H V Y T F V C K M P Q C	74
CD39Human.seq	T G V V H O V E E C R V K G P G I S K F V O K V N E I G I Y I T D C M E R A R E	114
246 prot	L P T L E G E V F D S I V K - P G L S A F V D Q P K Q G A E T V Q C L L E V A K D	113
CD39Human.seq	V I P R S Q H Q E T P V Y L G A T A G M R L L E M E S E E L A D R V L D V V E R	154
246 prot	S I P R S H W K K T P V V L K A T A G L R L L - - - P E H K A K A L L F E V K E	150
CD39Human.seq	S L S N Y P F D F Q - - G A R I I T G Q E E G A Y G W I T I N Y L L G K F S C K	192
246 prot	I F R K S I P F L V P K G S V S I M D G S D E G I L A W V T V N F I L T G Q C - - -	187
CD39Human.seq	T E W F S I V P Y E T N N Q E T F G A L D L G G A S T Q V T F V P Q - N Q T I E	231
246 prot	- - - - - H G H R Q E T V G T L D L G G A S T Q T F L P Q F E K T L E	218
CD39Human.seq	S P D N A - - L Q F R L Y G K D Y N V Y T H S E L C Y G K D O A L W C K L A K D	269
246 prot	Q T P R G Y L T S E E M F N S T Y K L Y T H S Y L G F G L K A A - - - R L A T L	255
CD39Human.seq	I O V A S N E I L R D P C F H P G Y K K V V N V S D L Y K T P C T K R - F E M T	308
246 prot	G A L E T E G - - - - - T D G H T F R L S A C L P R W L E A E	280
CD39Human.seq	L P F O O F - - - - - E I Q G I G N Y Q Q C H O S I L E L E N T S Y C P Y S Q	342
246 prot	W I E G G V K Y Q Y G G N Q E G E V G F E P C Y A E V L R V V R G K - - - - -	314
CD39Human.seq	C A F N G I F L P P L O G E F G A F S A F - - Y F V M K F L N L T S E K V S Q E	380
246 prot	- - - - - L H Q I D E E V Q R G S E Y A F S Y Y Y D R - - - A V D T D M I D Y E	345
CD39Human.seq	K V T E M - M K K F C A O P W E - - E I K T S Y A G V K E K Y L S E Y C F S G T	417
246 prot	K G G I L K V E D E F E R K A R E V C D N L E N F T S G S P - F L - - - C M D L S	381
CD39Human.seq	Y I L S L L L Q G Y R F T A D S W E H I H F I G K I Q G S D A G W T L G Y M L N	457
246 prot	Y I T A L L K D G F G F A D S T - - V L Q L T K K V N N I E T G W A L G A T F H	419
CD39Human.seq	L T N M I P A E C P L S T P L S H S T Y V F L M V L F S L V L F T V A I I G L L	497
246 prot	L L Q S L G I S H	428
CD39Human.seq	I F H K P S Y F W K D M I	510
246 prot		428

Figure 3

1	M A T S W G T V F F M L V V S C V I C S A V S H R N Q Q T W F E G I F L S S M C P	246	prot
1	M A T S W G A V F - M L I I A C V G S T V F Y R E Q Q T W F E G V F L S S M C P	mur	ntpase
41	I N V S A S T Y G I M F D A G S T G T R I H V Y T F V Q K M P I G Q L P I L E Q	246	prot
40	I N V S A G T F Y G I M F D A G S T G T R I H V Y T F V Q K T A G Q L P F L E Q	mur	ntpase
81	E V F D S V K P G L S A F V D Q P K Q G A E T V Q G L L E V A K D S I P R S H W	246	prot
80	E I F D S V K P G L S A F V D Q P K Q G A E T V Q E L L E V A K D S I P R S H W	mur	ntpase
121	K K T P V V L K A T A G L R L L P E H K A K A L L F E V K E I F R K S P F L V P	246	prot
120	E R T P V V L K A T A G L R L L P E Q K A Q A L L L E V E E I F K N S P F L V P	mur	ntpase
161	K G S V S I M D G S D E G I L A W V T V N F L T G Q L H G H R Q E T V G T L D L	246	prot
50	D G S V S I M D G S Y E G I L A W V T V N F L T G Q L H G R G Q E T V G T L D L	mur	ntpase
201	G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T Y K L Y T H	246	prot
200	G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T F K L Y T H	mur	ntpase
241	S Y L G F G L K A A R L A T L G A L E T E G T D G H T F R S A C L P R W L E A E	246	prot
240	S Y L G F G L K A A R L A T L G A L E A K G T D G H T F R S A C L P R W L E A E	mur	ntpase
281	W I F G G V K Y Q Y G G N Q E G E V G F E P C Y A E V L R V V R G K L H Q P E E	246	prot
280	W I F G G V K Y Q Y G G N Q E G E M G F E P C Y A E V L R V V Q G K L H Q P E E	mur	ntpase
321	V Q R G S F Y A F S Y Y Y D R A V D T D M I D Y E K G G I L K V E D F E R K A R	246	prot
320	V R G S A F Y A F S Y Y Y D R A A D T H L I D Y E K G G V L K V E D F E R K A R	mur	ntpase
361	E V C D N L E N F T S G S P F L C M D L S Y I T A L L K D G F G F A D S T V L Q	246	prot
360	E V C D N L G S F S S G S P F L C M D L T Y I T A L L K D G L G F A E R H P L T	mur	ntpase
401	L T K K V N N I E T G W - A L G A T F - - - - - H L L O S L G I S H	246	prot
400	- A H K E S E Q H R D W L G L G G H L S P A P V S G H H Q L R P S S T S E A C I	mur	ntpase
428		246	prot
439	S E P V F S O E G V D S E T F S D L S G K A W P E T R	mur	ntpase

Figure 4

Apyrase Conserved Regions in CD39-L4

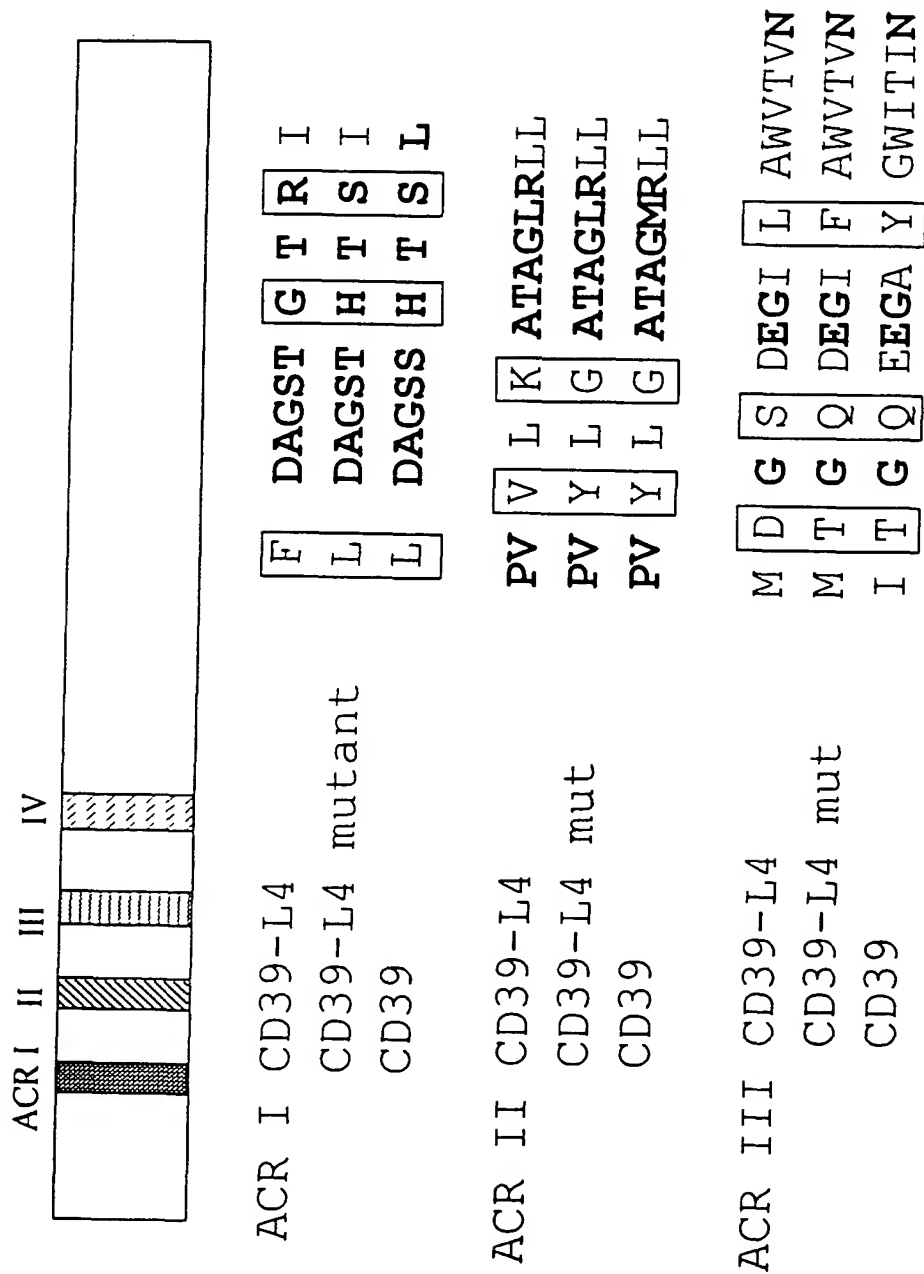


FIG. 5

Nucleotide sequence of the CD39-L4 mutant ACRIII (SEQ ID NO:6). The nucleotide changes have been highlighted. The G to A and A to C changes at positions 502 and 503 produce a Thr, the T to C, C to A and C to A changes at positions 508-510 result in a Gln and the A to C changed at position 525 result in a Phe.

ATGGCCACTTCTTGGGGCACAGTCTTTTTCATGCTGGTGGTATCCTGTGTTTGCAGCGCTGTCT
CCCACAGGAACCAGCAGACTTGGTTTGAGGGTATCTTCTGTCTTCCATGTGCCCCATCAATGT
CAGCGCCAGCACCTTGTATGGAATTATGTTTGATGCAGGGAGCACTGGAACCTCGAATTCATGTT
TACACCTTTGTGCAGAAAATGCCAGGACAGCTTCCAATTCTAGAAGGGGAAGTTTTTGATTCTG
TGAAGCCAGGACTTTTCTGCTTTTGTAGATCAACCTAAGCAGGGTGCTGAGACCGTTCAAGGGCT
CTTAGAGGTGGCCAAAGACTCAATCCCCCGAAGTCACTGGAAAAAGACCCAGTGGTCCTAAAG
GCAACAGCAGGACTACGCTTACTGCCAGAACACAAAGCCAAGGCTCTGCTCTTTGAGGTAAGG
AGATCTTCAGGAAGTCACCTTTCTGGTACCAAAGGGCAGTGTTAGCATCATG**ACTGGACAAGA**
CGAAGGCATATT**CGCTTGGGTT**ACTGTGAATTTTCTGACAGGTCAGTGCATGGCCACAGACAG
GAGACTGTGGGGACCTTGGACCTAGGGGGAGCCTCCACCCAAATCACGTTCTTGGCCCCAGTTTG
AGAAAACCTCTGGAACAACTCCTAGGGGCTACCTCACTTCCTTTGAGATGTTTAACAGCACTTA
TAAGCTCTATACACATAGTTACCTGGGATTGATTGAAAGCTGCAAGACTAGCAACCCTGGGA
GCCCTGGAGACAGAAGGGACTGATGGGCACACTTTCCGGAGTGCCTGTTTACCGAGATGGTTGG
AAGCAGAGTGATCTTTGGGGGTGTGAAATACCAGTATGGTGGCAACCAAGAAGGGGAGGTGGG
CTTTGAGCCCTGCTATGCCGAAGTGCTGAGGGTGGTACGAGGAAAACCTCACCAGCCAGAGGAG
GTCCAGAGAGGTTCTTCTATGCTTTCTTACTATTATGACCGAGCTGTTGACACAGACATGA
TTGATTATGAAAAGGGGGGTATTTTAAAGTTGAAGATTTTGAAGAAAAGCCAGGGAAGTGTG
TGATAACTTGGAACCTTCACC TCAGGCAGTCCTTTCTGTGCATGGATCTCAGCTACATCAC
AGCCCTGTTA AAGGATGGCTTTGGCTTTGCAGACAGCACAGTCTTACAGCTCACAAAGAAAGT
GAAC AACATAG AGACGGGCTGGGCCTTGGGGGCCACCTTTACCTGTTGCAGTCTCTGGGCA
TCTCCCATGA

Amino acid sequence of CD39-L4 mutant ACR III (SEQ ID NO:7). The amino acid changes are D to T (a.a. 168), S to Q (a.a. 170), and L to F (a.a. 175). The changes are shown in bold.

MATSYGTVFFMLVVSVCVSAVSHRNQQTWFEGIFLSSMCPINVSASTLYGIMFDAGSTGT
RIHVYTFVQKMPGQLPILEGEVFDVSKPGLSAFVDQPKQGAETVQGLLEVAKDSIPRSHW
KKTPVVLKATAGLRLLEPHKAKALLFEVKEIFRKSPFLVPKGSVSIM**TGQDEGIF**AWVTV
NFLTGQLHGHREQETVGTLDLGGASTQITFLPQFEKTLEQTPRGYLTSFEMFNSTYKLYTH
SYLGFGFLKAARLATLGALETEGTDGHTFRSACLPRWLEAEWIFGGVKYQYGGNQEGEVGF
EPCYAEVLRVVRGKLHQPEEVQRGSFYAFSYYYDRAVDTDMIDYEKGGILKVEDFERKAR
EVCNLENFTSGSPFLCMDLSYITALLKDGFGFADSTVLQLTKKVNNIETGWALGATFHL
LQSLGISH

FIG. 6

**FIG. 2 ADPase activity of CD39-L4
 ACR mutants**



1) ACR I; 2) ACR II; 3) ACR III; 4)
 Wild Type; 5) CD39; 6) Vector

CD39-L4

FIG. 7